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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.
08/876,276	06/16/97	SHORT J	09010/020001

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EXAMINER	
TUNG, P	
ART UNIT	**PAPER NUMBER**
1652	11

DATE MAILED: 12/07/99

Please find below and/or attached an Office communication concerning this application or proceeding.

Commissioner of Patents and Trademarks

Office Action Summary	Application No. 08/876,276	Applicant(s) Short et al.
	Examiner Peter Tung	Group Art Unit 1652

☐ Responsive to communication(s) filed on _____.

☐ This action is **FINAL.**

☐ Since this application is in condition for allowance except for formal matters, **prosecution as to the merits is closed** in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11; 453 O.G. 213.

A shortened statutory period for response to this action is set to expire 3 month(s), or thirty days, whichever is longer, from the mailing date of this communication. Failure to respond within the period for response will cause the application to become abandoned. (35 U.S.C. § 133). Extensions of time may be obtained under the provisions of 37 CFR 1.136(a).

Disposition of Claims

☒ Claim(s) *1-18* is/are pending in the application.

Of the above, claim(s) _____ is/are withdrawn from consideration.

☐ Claim(s) _____ is/are allowed.

☒ Claim(s) *1-18* is/are rejected.

☐ Claim(s) _____ is/are objected to.

☐ Claims _____ are subject to restriction or election requirement.

Application Papers

☐ See the attached Notice of Draftsperson's Patent Drawing Review, PTO-948.

☐ The drawing(s) filed on _____ is/are objected to by the Examiner.

☐ The proposed drawing correction, filed on _____ is ☐approved ☐disapproved.

☐ The specification is objected to by the Examiner.

☐ The oath or declaration is objected to by the Examiner.

Priority under 35 U.S.C. § 119

☐ Acknowledgement is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d).

☐ All ☐ Some* ☐ None of the CERTIFIED copies of the priority documents have been

☐ received.

☐ received in Application No. (Series Code/Serial Number) _____.

☐ received in this national stage application from the International Bureau (PCT Rule 17.2(a)).

*Certified copies not received: _____.

☐ Acknowledgement is made of a claim for domestic priority under 35 U.S.C. § 119(e).

Attachment(s)

☒ Notice of References Cited, PTO-892

☒ Information Disclosure Statement(s), PTO-1449, Paper No(s). 6

☐ Interview Summary, PTO-413

☐ Notice of Draftsperson's Patent Drawing Review, PTO-948

☐ Notice of Informal Patent Application, PTO-152

--- SEE OFFICE ACTION ON THE FOLLOWING PAGES ---

DETAILED ACTION

Election/Restriction

1. Applicant's election with traverse of esterases and acidophiles in Paper No. 9, dated 8/30/99 is acknowledged. The traversal is on the ground(s) that the invention is useful for identification of any bioactivity in any prokaryote and that the species of enzymes are not patentably distinct as it is the method itself that is patentable and applicable to any bioactivity. Additionally, the method is equally applicable to any species listed in claim 10. An election of species is no longer required as Applicants have stated that the instant method is applicable to any bioactivity and species listed in claim 10. Applicants have also stated that the species of enzymes and organisms are not patentably distinct as it is the method that is patentable.

Specification

2. This application contains sequence disclosures that are encompassed by the definitions for nucleotide and/or amino acid sequences set forth in 37 CFR 1.821(a)(1) and (a)(2). However, this application fails to comply with the requirements of 37 CFR 1.821 through 1.825 for the following reason(s): sequences are disclosed on page 69, lines 11 and 13 which are not included in a Sequence Listing or in computer readable form.

Claim Objections

3. Claim 11 is objected to because of the following informalities: What "C12FDG" represents should be spelled out. Appropriate correction is required.

4. Claim 16 is objected to because of the following informalities: What "FACS" represents should be spelled out. Appropriate correction is required.

5. Claim 12 is objected to under 37 CFR 1.75(c), as being of improper dependent form for failing to further limit the subject matter of a previous claim. Applicant is required to cancel the claim(s), or amend the claim(s) to place the claim(s) in proper dependent form, or rewrite the claim(s) in independent form. Claim 10, the claim prior to claim 12, is drawn to a method using extremophiles and not to substrates.

Claim Rejections - 35 USC § 112

6. The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

7. Claims 1-18 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

8. Claim 1 is indefinite as "sample(s)" in the claim is used in referring to DNA (genomic DNA samples, DNA sequence of positive samples) as well as a host cell (DNA of said samples, substrates into samples).

9. The term "normalized" in claim 1 is a relative term which renders the claim indefinite. The term "normalized" is not defined by the claim, the specification does not provide a standard for ascertaining the requisite degree, and one of ordinary skill in the art would not be reasonably apprised of the scope of the invention. For the purposes of this Office action, "normalized" is understood to mean DNA which has undergone the normalization procedure described on page 69, line 20 to page 70, line 6.

10. The term "multispecific" in claim 1 is a relative term which renders the claim indefinite. The term "multispecific" is not defined by the claim, the specification does not provide a standard for ascertaining the requisite degree, and one of ordinary skill in the art would not be reasonably apprised of the scope of the invention. It is unclear what a multispecific expression library would be.

11. Claim 11 is unclear as to what would comprise a bioactive substrate as only C12FDG is listed. Changing "comprises" to "is" would correct the problem.

12. Claim 14 and 15 are indefinite as a single temperature of about 70°C or a single time of about 30 minutes, respectively, is not a range.

13. Claim 18 is indefinite as it is unclear as to what comprises the method of the instant claim. The claim is unclear as it refers to the method of claim 1 but does not make clear what the instant method is supposed to do.

14. Claims 2-18 are indefinite because they depend upon an indefinite base claim and fail to correct the problem.

15. The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

16. Claim 2 is rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for a method of screening DNA to identify lipases, esterases, glycosidases, proteases and monooxygenases through the use of bioactive fluorescent substrates does not reasonably provide enablement for the use of said screening method to identify glycosyl transferases, phosphatases, kinases, diarylpropane peroxidases, epoxide hydrolases, nitrile hydratases, nitrilases, transaminases, amidases and acylases. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the invention commensurate in scope with these claims. Enablement requires that the specification teach those in the art to make and use the invention without undue experimentation. Factors to be considered in determining whether a disclosure would require undue experimentation include (1) the quantity of experimentation necessary, (2) the amount of direction or guidance presented, (3) the presence or absence of working examples, (4) the nature

of the invention, (5) the state of the prior art, (6) the relative skill of those in the art, (7) the predictability or unpredictability of the art, and (8) the breadth of the claims. The breadth of the claims encompasses the assay of several enzymes recited in the instant claim by using fluorescent substrates. However, insufficient examples are provided of fluorescent substrates which can be used to assay enzymes besides lipases, esterases, glycosidases, proteases and monooxygenases. A large amount of experimentation would be required to determine which fluorescent substrates would be useful in assaying the other enzymes recited in the claim. The relative skill of those in the art is low in determining which fluorescent substrates can be catalyzed by a particular enzyme and used as an indicator of enzymatic activity. There is unpredictability in the art in determining fluorescent substrates which are useful as indicators of enzymatic activity. Insufficient guidance is provided on identifying and/or making fluorescent substrates for the enzymes besides lipases, esterases, glycosidases, proteases and monooxygenases. Given the limiting scope of the disclosure, undue experimentation would be required to enable the full scope of the claim.

Claim Rejections - 35 USC § 102

17. The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless --
>
> (e) the invention was described in a patent granted on an application for patent by another filed in the United States before the invention thereof by the applicant for patent, or on an international application by another who has fulfilled the requirements of paragraphs (1), (2), and (4) of section 371(c) of this title before the invention thereof by the applicant for patent.

18. Claims 1, 4-10, 16 is rejected under 35 U.S.C. 102(e) as being anticipated by Thompson et al. (U.S. Patent No. 5,824,485). Thompson et al. teach (column 46, line 61 to column 47, line 50; column 37, lines 29-36; column 31, line 58 to column 32, line 56; column 6, lines 36-41) a method of screening a normalized (biased) expression library by screening for the presence of a desired activity. Thompson et al. teach (column 33, lines 28-41; column 35, lines 10-19; column 36, lines 60-65) that activity can be detected by fluorescence activated cell sorting when a fluorescent probe is used, substrates labeled with a fluorogenic agent can serve as probes of specific enzymatic activity in cells upon incubation with the fluorescent substrate (column 36, line 66 to column 37, line 25), and the genes of a biochemical pathway identified by screening an expression library can then be sequenced. Thompson et al. also teach (column 37, lines 37-54) FACS of gel encapsulated samples. Additionally, Thompson et al. teach (column 15, Table I; column 14, lines 1-3) that DNA donors for the expression library may be gram negative prokaryotes, thermophiles, halophiles, or acidophiles. The instant claims are therefore anticipated by Thompson et al.

Claim Rejections - 35 USC § 103

19. The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person

having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

20. This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. 103(a), the examiner presumes that the subject matter of the various claims was commonly owned at the time any inventions covered therein were made absent any evidence to the contrary. Applicant is advised of the obligation under 37 CFR 1.56 to point out the inventor and invention dates of each claim that was not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of 35 U.S.C. 103(c) and potential 35 U.S.C. 102(f) or (g) prior art under 35 U.S.C. 103(a).

21. Claims 1 and 2 are rejected under 35 U.S.C. 103(a) as being unpatentable over Thompson et al. (U.S. Patent No. 5,824,485) in view of Nader et al.(U.S. Patent No. 5,173,187). The teachings of Thompson et al. as they apply to claim 1 have been discussed supra. Thompson et al. do not teach screening an expression library for esterase activity. Nader et al. teach the identification of esterases in bacteria by their cleavage of a fluorescent substrate and the isolation of the bacteria by fluorescence activated cell sorting. Nader et al. do not teach screening an expression library for esterase activity. It would have been obvious to one of ordinary skill in the art at the time the invention was made to screen an expression library, as taught by Thompson et al., for esterase activity using a fluorescent substrate, as taught by Nader et al. One of ordinary skill in the art is motivated to do this for the benefit of identifying a clone of the expression library which expresses an esterase. One of ordinary skill in the art would have a reasonable expectation of success at doing this as the teachings of Nader et al. show that bacteria expressing esterases

can be identified and isolated by fluorescence activated cell sorting of cells which cleave an fluorescent esterase substrate. It would be a reasonable expectation that this fluorescent substrate could be then used to screen for esterases in the method of screening an expression library as shown by the teachings of Thompson et al. Therefore the invention as a whole would have been prima facie obvious to a person of ordinary skill in the art at the time the invention was made.

22. Claims 1 and 3 are rejected under 35 U.S.C. 103(a) as being unpatentable over Thompson et al. (U.S. Patent No. 5,824,485). Thompson et al. qualifies as prior art under 35 U.S.C 102(e) as it has an effective filing date of 4/25/95. The teachings of Thompson et al. as they apply to claim 1 have been discussed supra. Claim 3 adds the further limitation of the prokaryotic expression library containing at least of about 2×10^6 clones. Although Thompson et al. do not teach the use of at least 2×10^6 clones in the expression library, this claim limitation is dependent upon the size of the DNA of the donor organism used to generate an expression library. One of ordinary skill in the art would produce an expression library containing a sufficient number of clones in order to have a statistically sufficient probability of having all desired DNA sequences of the donor organism represented by the expression library. One of ordinary skill in the art would have a reasonable expectation of success at doing this as the requirement of having a sufficient number of clones in a DNA library in order to include desired DNA sequences is well known in the art. Therefore the invention as a whole would have been prima facie obvious to a person of ordinary skill in the art at the time the invention was made.

23. Claims 1, 3, 10, 11 and 12 are rejected under 35 U.S.C. 103(a) as being unpatentable over Thompson et al. (U.S. Patent No. 5,824,485) in view of Miao et al. Thompson et al. qualifies as prior art under 35 U.S.C 102(e) as it has an effective filing date of 4/25/95. The teachings of Thompson et al. as they apply to claims 1, 3 and 10 have been discussed supra. Claim 11 adds the further limitation of a C12FDG substrate and claim 12 adds the further limitation of a substrate comprising a lipophilic tail. Thompson et al. do not teach C12FDG or a substrate comprising a lipophilic tail. Miao et al. teach ("Abstract,"page 708) assaying *E. coli* for β-galactosidase activity by fluorescence flow cytometry using the fluorescent substrate C12FDG. Miao et al. do not teach a method of screening an expression library using C12FDG. It would have been obvious to one of ordinary skill in the art at the time the invention was made to screen an expression library, as taught by Thompson et al., using C12FDG, as taught by Miao et al. for the benefit of FACS screening an expression library for β-galactosidase activity. One of ordinary skill in the art is motivated to combine the two references as Miao teaches that bacteria with β-galactosidase activity can be identified by fluorescence flow cytometry using the substrate C12FDG. As fluorescence flow cytometry is the basis for detection in FACS and Thompson et al. teach screening of an expression library using FACS and fluorescent substrates, one of ordinary skill in the art would have used the β-galactosidase substrate C12FDG in a FACS screening of an expression library. One of ordinary skill in the art would have a reasonable expectation of success at doing this as it would be a reasonable expectation to be able to use FACS screening of an expression library, as shown by the teachings of Thompson et al., with the fluorescent β-

galactosidase substrate C12FDG, as shown by the teachings of Miao et al., to isolate β-galactosidase expressing clones as the teachings of Miao et al. show using fluorescence flow cytometry analysis of *E. coli* using C12FDG. Additionally, C12FDG comprises a lipophilic tail. Therefore the invention as a whole would have been prima facie obvious to a person of ordinary skill in the art at the time the invention was made.

24. Claims 1 and 17 are rejected under 35 U.S.C. 103(a) as being unpatentable over Thompson et al. (U.S. Patent No. 5,824,485). Thompson et al. qualifies as prior art under 35 U.S.C 102(e) as it has an effective filing date of 4/25/95. The teachings of Thompson et al. as they apply to claim 1 have been discussed supra. Claim 17 adds the further limitation of biopanning the prokaryotic expression library before inserting bioactive substrates. Thompson et al. further teach (column 41, lines 25-50) using subtracted DNA probes to prescreen an expression library . It would have been obvious to one of ordinary skill in the art at the time the invention was made to prescreen an expression library using DNA probes and to then screen selected clones by fluorescence activated cell sorting. One of ordinary skill in the art is motivated to do this for the benefit of screening a library containing clones enriched for specific genes. One of ordinary skill in the art has a reasonable expectation of success at doing this as teachings of Thompson et al. show using DNA probes for prescreening a library. It would be a reasonable expectation that this prescreening method can be used with the FACS method of screening an expression library as shown by the teachings of Thompson et al. Therefore the invention as a

whole would have been prima facie obvious to a person of ordinary skill in the art at the time the invention was made.

25. Claims 1 and 13-15 are rejected under 35 U.S.C. 103(a) as being unpatentable over Thompson et al. (U.S. Patent No. 5,824,485) in view of Miao et al. Thompson et al. qualifies as prior art under 35 U.S.C 102(e) as it has an effective filing date of 4/25/95. The teachings of Thompson et al. as they apply to claim 1 have been discussed supra. Claims 13-15 add the further limitation of a heating samples, heating samples at about 70°C, and heating for about 30 minutes, respectively. Thompson et al. do not teach heating samples before screening an expression library. Miao et al. teach ("Abstract,"page 708; "Single-Cell β-Galactosidase Assay, page 709) assaying *E. coli* for β-galactosidase activity by fluorescence flow cytometry using the fluorescent substrate C12FDG and growing E. coli cells at 37°C for a few minutes to an hour to allow permeation and reaction of the fluorescent substrate. Miao et al. do not teach a method of screening an expression library or heating cells at 70°C. It would have been obvious to one of ordinary skill in the art at the time the invention was made to screen an expression library, as taught by Thompson et al., by heating cells for a few minutes to an hour to allow permeation and reaction of the fluorescent substrate as taught by Miao et al. for the benefit of FACS screening an expression library for β-galactosidase activity. It would have been obvious to one of ordinary skill in the art to heat the samples at a temperature required for activity of the enzymes being screened. Elevated temperatures would be required for enzymes from thermophilic sources. One of ordinary skill in the art is motivated to combine the two references as Miao teaches that

bacteria with β-galactosidase activity can be identified by fluorescence flow cytometry using the substrate C12FDG. As fluorescence flow cytometry is the basis for detection in FACS and Thompson et al. teach screening of an expression library using FACS and fluorescent substrates, one of ordinary skill in the art would have used the β-galactosidase substrate C12FDG in a FACS screening of an expression library. One of ordinary skill in the art would have a reasonable expectation of success at doing this as it would be a reasonable expectation to be able to use FACS screening of an expression library, as shown by the teachings of Thompson et al., with the fluorescent β-galactosidase substrate C12FDG, as shown by the teachings of Miao et al., to isolate β-galactosidase expressing clones as the teachings of Miao et al. show using fluorescence flow cytometry analysis of *E. coli* using C12FDG. The teachings of Miao et al. show that heating of samples is required for permeation and reaction of the fluorescent substrate. Therefore the invention as a whole would have been prima facie obvious to a person of ordinary skill in the art at the time the invention was made.

Allowable Subject Matter

26. Claim 18 is allowable over the prior art of record.

27. Claim 18 would be allowable if rewritten to overcome the rejection(s) under 35 U.S.C. 112, 2nd paragraph, set forth in this Office action and to include all of the limitations of the base claim and any intervening claims.

28. No claims are allowed.

29. Any inquiry concerning this communication or earlier communications from the examiner should be directed to Peter Tung, Ph.D. whose telephone number is (703) 308-9436. The examiner can normally be reached on Monday-Friday from 9:00 to 5:30.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Ponnathapu Achutamurthy, Ph.D., can be reached on (703) 308-3804. The fax phone number for the organization where this application or proceeding is assigned is (703) 308-0294.

Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to the receptionist whose telephone number is (703) 308-0196.

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